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8-7-02

Docket No.: HYS-28

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s):	Boyle, et al.
Serial No:	09/676,135
Filed:	September 29, 2000
For:	METHODS AND MATERIALS RELATING TO METALLOCARBOXYPEPTIDASE-LIKE POLYPEPTIDES AND POLYNUCLEOTIDES
Examiner:	Jehanne E. Souaya
Group:	1634

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AMENDMENT AND RESPONSE UNDER
37 CFR § 1.113 TO OFFICE ACTION MAILED JUNE 19, 2002

This is in response to the Office Action mailed June 19, 2002, which is due on or before September 19, 2002. No Extension of Time is required to file this response. In the Office Action, the Examiner rejected claims 10-13, 24, 25 and 29. Applicants traverse the Examiner's rejections as set forth below, and request that the Examiner consider the following remarks, which Applicants respectfully submit demonstrate that the claims are in condition for allowance. The Commissioner is hereby authorized to charge any additional fees necessary in connection with this paper to Deposit Account No. 50-1169.

In the Office Action dated June 19, 2002, the Examiner rejected claims 10-13, 24, 25 and 29 under 35 U.S.C. 101, 112, 1st and 2nd and 102. Applicants respectfully request reconsideration in light of the following amendments and remarks.

AMENDMENTS

In the specification:

Please replace the paragraph at page 116, line 3 with the following amended paragraph at page 116, line 3:

B1

A polypeptide was predicted to be encoded by SEQ ID NO: 2 as set forth below. The polypeptide was predicted using a software program called FASTY (available from the Internet website at: fasta.bioch.virginia.edu) which selects a polypeptide based on a comparison of translated novel polynucleotide to known polypeptides (W.R. Pearson, Methods in Enzymology, 183: 63-98 (1990), herein incorporated by reference).

In the Claims:

Please cancel claims 12-13.

Please add the following new claim.

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30. An isolated polypeptide consisting of an amino acid sequence which is 99% identical to the amino acid sequence of SEQ ID NO: 4.

REMARKS

Upon entry of this amendment, claims 10, 11, 25, and 30 are pending in the application and are set forth in **Appendix B** hereto. A marked up version of the amended specification is attached as **Appendix A** hereto.

In this response, claims 1-9, 12-23, and 26-28 have been canceled, and claim 30 has been added. Support for new claim 30 can be found in the specification at page 37, lines 5-13. No new matter is added.

Examiner's Position

In the Office Action dated June 19, 2002, the Examiner made the following rejections:

(1) Claims 10-13, and 25 were rejected under 35 U.S.C. §101 as not supported by a specific, substantial, credible, or well-established utility;

(2) Claims 10-13, and 25 were rejected under 35 U.S.C. §112, first paragraph, as subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention;

(3) Claims 12 and 13 were rejected under 35 U.S.C. §112, first paragraph, for lack of written description;

(4) Claim 13 was rejected under 35 U.S.C. §112, second paragraph, for being indefinite; and

(5) Claim 13 was rejected under 35 U.S.C. §102, as being anticipated by Dumas.

Applicants traverse each of these rejections as follows.

35 U.S.C. §101 Utility Rejection Should Be Withdrawn

The Examiner alleged, "Claims 10-13 and 25 are rejected under 35 U.S.C.§ 101, because the claimed invention is not supported by either a specific or substantial asserted utility or a well established utility..." (Office action at p. 4.) The Applicants respectfully traverse.

Applicants submit that the specification contains clear assertions of a well-established utility for the polypeptide of the invention. As disclosed in the specification at page 2, lines 12-14; and page 4, lines 13-20, the claimed polypeptide is homologous to members of the metallocarboxypeptidase (metallo-CP) family, which have a well-established enzymatic utility.

Page 117, lines 19-23, of the specification states that SEQ ID NO: 4 recited in the claims shares homology with human carboxypeptidase B (a metallocarboxypeptidase). One of ordinary skill in the art accepts structural homology based on amino acid sequence identity as a credible method of determining the function of a polypeptide. *See* Henikoff et al., Science, 278:609-614 (1997). In addition, page 118, lines 15-17; and page 118, line 25 – page 120, line 15 of the specification also describe the presence of motifs and signature domains, **all** consistent with SEQ

ID NO: 4 having carboxypeptidase, and in particular, metallocarboxypeptidase, activity and being a member of the carboxypeptidase family. Reznik and Fricker, *Cell. Mol. Life. Sci.*, 58:1790-1804, 1790 (2001). Although SEQ ID NO: 4 shares the highest homology with other members of this family in its catalytic domain, homology over this domain is highly significant for predicting activity. *Id.* at 1791, Fig. 1; Vendrell et al., *Biochim. Biophys. Acta.*, 1477:284-298, 285 (2000); NiceSite View of PROSITE: PS00132; and Pfam 7.4 (Saint Louis) PF00246: Zinc carboxypeptidase.

The metallo-CP family can be generally broken down into two sub-families in accordance with the function of the molecule. It is well known in the art that within the metallo-CP family, there are only two possible biological functions exists for all members, including plants and bacteria, namely digestive and regulatory. Wei at 14954; Skidgel and Erdos, *Immunological Reviews*, 161:129-141, 130 (1998); Vendrell at 285; Reznik at 1790; and Pfam at 1. The recognized and well-established sequence homology between sub-families is approximately 20%, while homology within a sub-family is generally over 40%. Wei et al., *J. Biol. Chem.*, 277(17): 14954-14964, 14954-55 (2002); Reznik at Fig. 1; and Vendrell at 285. As demonstrated in Figures 1 & 2 of the application, the claimed polypeptide shares a 46% and 48% homology with Bothrops jararaca metallo-CP and human metallo-CP B mutant respectively, which falls within the expected degree of homology for sub-family members. The distinction, therefore, between carboxypeptidase families is based on the overall domain structure, as well as the amino acid sequence similarities of these genes. Wei at 14954.

Applicants submit herewith bioinformatic evidence in the form of a Clustal W analysis that further demonstrates that the polypeptide of SEQ ID NO: 4 is a digestive type metallocarboxypeptidase, because: (1) it exhibits homology above the expected average for sub-family members; (2) it contains all the signature domains, and conserved amino acid residues; and (3) it does not contain a transthyretin-like folding domain present in all metallocarboxypeptidase regulatory type family members. (See Appendix **C**) In Appendix **C** the claimed polypeptide is aligned with human CPO (a member of the metallo-CP digestive sub-family); pig prometallo-CP-B; mouse thrombin carboxypeptidase(a member of the metallo-CP regulatory sub-family); and mouse carboxypeptidase R(a member of the metallo-CP regulatory sub-family). The digestive sub-family is referred to as the A/B metallo-CP subfamily, and all

members contain the HXXE...H zinc binding motif, where the three zinc ligands are a histidine, a glutamate located two residues downstream, and a second histidine located further downstream from the first histidine. Vendrell at 285; and Pfam at 1. This motif is identified by asterisks in the Clustal W alignment (Appendix C, residues 108, 111 and 236). In addition, PROSITE:PS00132 identified a pattern that is unique to the metallo-CPs, namely, [PK]x[LIVMFY]x[LIVMFY]x(4)H[STAG]xEx[LIVM][STAG]x(6)[LIVMFYTA], wherein, the bracketed groups represent one residue which can contain any of the amino acids listed, x is any amino acid, and a non-bracketed amino acid is essential residue. This pattern can be identified at residues K99-F121 of the claimed polypeptide in the Clustal W alignment. The digestive, or A/B metallo-CPs are distinguished from the regulatory sub-family members by the fact that **all** members of the regulatory sub-family contain a transthyretin-like folding domain that is immediately C-terminal to the active CP domain, which is absent in the claimed polypeptide, whereas all the digestive enzyme members contain an N-terminal pro-region, and one active carboxypeptidase (CP) domain ~300 amino acids in length. Wei at 14954; and Reznik at 1791. Finally, the active site and a substrate-binding domain of the A/B metallo-CP subfamily contain highly conserved residues, which are necessary to render the molecule active. Wei at 14955, 14959 and Fig. 2. The claimed polypeptide contains all of the conserved residues which are identified in the Clustal W alignment by a square atop the residue, namely residues 110, 163, 166, 167, 183, 184, 237, 238, 288, 310, and 319 of the polypeptide of SEQ ID NO: 4. Therefore, base on the fact that SEQ ID NO: 4 of the claimed invention exhibits homology above the expected average for metallo-CP sub-family members, and it contains all the signature domains, and conserved amino acid residues SEQ ID NO: 4 is a member of the digestive metallocarboxypeptidase subfamily, and thus has a well-established biological function and utility.

Preliminary experiments were carried out using supernatant containing the polypeptide of SEQ ID NO: 4. The results from these experiments were inconclusive; no statistically significant activity was observed on the substrates hippuryl-Arginine and hippuryl-Leucine-phenylalanine, and different results may be observed with other substrates. Bioinformatic evidence indicates the structural characteristics of SEQ ID NO: 4 is so closely align with those of known members of

the digestive-type metallo-CPs, that the claimed polypeptide is definitively a digestive metallocarboxypeptidase.

As such, this is a well-established utility because one of ordinary skill in the art would clearly understand from the knowledge in the art that metallocarboxypeptidase activity is useful in and of itself, and identification as a digestive metallo-CP would not require undue experimentation, as this family and subfamilies are so well defined and recognized in the art.

35 U.S.C. §112, First Paragraph, Rejections Should Be Withdrawn

In the Office Action, the Examiner rejected Claims 10-13 and 25 under 35 U.S.C. § 112, first paragraph, because one of ordinary skill in the art would allegedly not know how to use the invention since the claimed invention was said to lack utility. Applicants have disclosed the sequence for claimed subject matter. One of skill in the art would know how to make the claimed invention based on the disclosed sequence. As described above, the sequence of the pending claims has a well-established utility as a member of the metallocarboxypeptidase family of enzymes. Since the subjects of the pending claims have been demonstrated to have such utility, Applicants respectfully submit that they are properly enabled, and as such, the rejection is moot. Applicants therefore request that this §112 rejection be withdrawn.

Claims 12 and 13 Satisfy the Written Description Requirement

In the Office Action, the Examiner rejected Claims 12 and 13 under 35 U.S.C. § 112, first paragraph, because the subject matter described in the specification allegedly did not reasonably convey to one skilled in the art that applicants had possession of the claimed invention at the time the application was filed.

Specifically, the Examiner rejected claims 12 and 13 under 35 U.S.C. §112, first paragraph, for failing to meet the written description requirement. The Examiner maintains that there is not adequate written description for claims directed to a genus of polypeptides compromising the sequence of SEQ ID NO: 4 a portion thereof, at least 5 or at least 10 consecutive nucleotides thereof. On page 15, the Examiner acknowledged that the polypeptide sequence of SEQ ID NO: 4 complies with the written description requirements.

Without acceding to the propriety of the Examiner's position, and in order to facilitate prosecution, applicants have canceled claims 12 and 13. Accordingly, the rejection as applied to these claims is moot. Applicants believe that upon entry of this amendment, the §112 written description rejection of claims 12 and 13 has been overcome so that these claims should be found allowable. Withdrawal of the rejection is therefore respectfully requested.

Enablement Rejections are Overcome

On pages 9-15 of the Office Action, the Examiner rejected Claims 10-13 and 25 under 35 USC § 112, first paragraph, because the subject matter described in the specification allegedly did not enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention. The Examiner's position appears to be that it would require undue experimentation for a skilled artisan to make and use the polypeptide of SEQ ID NO: 4, nor variants or homologs of SEQ ID NO: 4, because "the specification does not define what activity is encompassed by the term 'carboxypeptidase-like.'"

Applicants respectfully traverse this rejection for the following reasons. As described in the preceding section of the present response, Applicants have demonstrated that an artisan skilled in the art would be able to make and use the polypeptide of SEQ ID NO: 4 as it contains all the signature sequences, distinctive domains, and highly conserved residues well known in the art to identify metallocarboxypeptidase family members, and digestive sub-family members without undue experimentation. In addition, the "activity" associated with carboxypeptidases are well established in the art and are not far reaching, but are confined to basic types, namely regulatory and digestive.

Without acceding to the propriety of the Examiner's position, and in order to facilitate prosecution, applicants have canceled claims 12 and 13 to variants and homologs. Accordingly, the rejection should be withdrawn.

Indefiniteness Rejection Overcome

On page 18 of the Office Action, the Examiner rejected claim 13 under 35 USC § 12, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the

subject matter which applicant regards as the invention. The Examiner's position is that the claims recite "the polynucleotide of SEQ ID NO: 4" and SEQ ID NO: is a polypeptide.

Without acceding to the propriety of the Examiner's position, and in order to facilitate prosecution, applicants have cancelled claim 13. Applicants believe that upon entry of this amendment, the § 112 indefiniteness rejection of claim 13 is moot. Withdrawal of the rejection is therefore respectfully requested.

35 USC § 102 Anticipation Rejections Overcome

On page 19 of the Office Action, the Examiner rejected claim 13 under 35 USC § 102(a) as being anticipated by Dumas, EP1033401, Sept. 6, 2000. Applicants respectfully traverse the rejections for the reasons discussed below.

Without acceding to the propriety of the Examiner's position, and in order to facilitate prosecution, applicants have cancelled claim 13. Applicants believe that upon entry of this amendment, the § 112 indefiniteness rejection of claim 13 is moot. Withdrawal of the rejection is therefore respectfully requested.

CONCLUSION

On the basis of the foregoing amendments and remarks, Applicants respectfully submit that the pending claims are in condition for allowance, and a Notice of Allowance is respectfully requested as soon as possible. If there are any questions regarding these amendments and

remarks, or if further discussion would expedite allowance of the claims, the Examiner is encouraged to contact the undersigned at the telephone number provided below.

Respectfully submitted,

Date: August 1, 2002

By: Elena Quertermous

Elena E. Quertermous, Ph.D.
Agent for Applicants
Registration No.: 47,873
HYSEQ, INC.
675Almanor Avenue
Sunnyvale, CA 94085
(408) 524-8100
(408) 524-8145 (Telefacsimile)